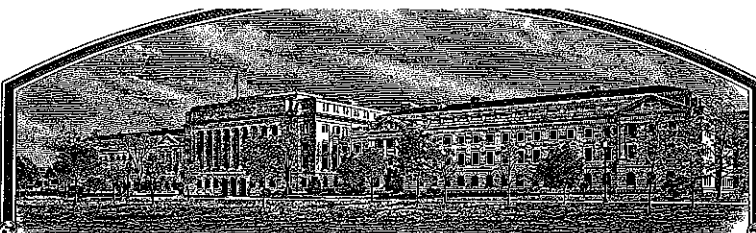


No.

200600022



THE UNITED STATES OF AMERICA

TO ALL TO WHOM THESE PRESENTS SHALL COME:

NASH Research Foundation

Whereas, THERE HAS BEEN PRESENTED TO THE

Secretary of Agriculture

AN APPLICATION REQUESTING A CERTIFICATE OF PROTECTION FOR AN ALLEGED DISTINCT VARIETY OF SEXUALLY REPRODUCED, OR TUBER PROPAGATED PLANT, THE NAME AND DESCRIPTION OF WHICH ARE CONTAINED IN THE APPLICATION AND EXHIBITS, A COPY OF WHICH IS HEREUNTO ANNEXED AND MADE A PART HEREOF, AND THE VARIOUS REQUIREMENTS OF LAW IN SUCH CASES MADE AND PROVIDED HAVE BEEN COMPLIED WITH, AND THE TITLE THERETO IS, FROM THE RECORDS OF THE PLANT VARIETY PROTECTION OFFICE, IN THE APPLICANT(S) INDICATED IN THE SAID COPY, AND WHEREAS, UPON DUE EXAMINATION MADE, THE SAID APPLICANT(S) IS (ARE) ADJUDGED TO BE ENTITLED TO A CERTIFICATE OF PLANT VARIETY PROTECTION UNDER THE LAW.

NOW, THEREFORE, THIS CERTIFICATE OF PLANT VARIETY PROTECTION IS TO GRANT UNTO THE SAID APPLICANT(S) AND THE SUCCESSORS, HEIRS OR ASSIGNS OF THE SAID APPLICANT(S) FOR THE TERM OF TWENTY YEARS FROM THE DATE OF THIS GRANT, SUBJECT TO THE PAYMENT OF THE REQUIRED FEES AND PERIODIC REPLENISHMENT OF VIABLE BASIC SEED OF THE VARIETY IN A PUBLIC REPOSITORY AS PROVIDED BY LAW, THE RIGHT TO EXCLUDE OTHERS FROM SELLING THE VARIETY, OR OFFERING IT FOR SALE, OR REPRODUCING IT, OR SELLING IT, OR EXPORTING IT, OR CONDITIONING IT FOR PROPAGATION, OR STOCKING IT FOR ANY OF THE FOREGOING PURPOSES, OR USING IT IN PRODUCING A HYBRID OR DIFFERENT VARIETY THEREFROM, TO THE EXTENT PROVIDED BY THE PLANT VARIETY PROTECTION ACT. (84 STAT. 1542, AS AMENDED, 7 U.S.C. 2321 ET SEQ.)

OAT

'STARK'

In Testimony Whereof, I have hereunto set my hand and caused the seal of the Plant Variety Protection Office to be affixed at the City of Washington, D.C. this sixth day of December, in the year two thousand and six.

Attest:

Commissioner
Plant Variety Protection Office
Agricultural Marketing Service

Secretary of Agriculture

U.S. DEPARTMENT OF AGRICULTURE
AGRICULTURAL MARKETING SERVICE
SCIENCE AND TECHNOLOGY - PLANT VARIETY PROTECTION OFFICE

APPLICATION FOR PLANT VARIETY PROTECTION CERTIFICATE
(Instructions and information collection burden statement on reverse)

The following statements are made in accordance with the Privacy Act of 1974 (5 U.S.C. 552a) and the Paperwork Reduction Act (PRA) of 1995.

Application is required in order to determine if a plant variety protection certificate is to be issued (7 U.S.C. 2421). Information is held confidential until certificate is issued (7 U.S.C. 2426).

1. NAME OF OWNER NDSU Research Foundation		2. TEMPORARY DESIGNATION OR EXPERIMENTAL NAME ND960736		3. VARIETY NAME 'Stark'	
4. ADDRESS (Street and No., or R.F.D. No., City, State, and ZIP Code, and Country) C/O Executive Director 1735 NDSU Research Park Drive PO Box 5002 Fargo, ND 58105-5002		5. TELEPHONE (include area code) 701-231-8931		FOR OFFICIAL USE ONLY PVP NUMBER 200600022 FILING DATE October 28, 2005	
		6. FAX (include area code) 701-231-6661			
7. IF THE OWNER NAMED IS NOT A "PERSON", GIVE FORM OF ORGANIZATION (corporation, partnership, association, etc.) NDSU Research Foundation 501(c)(3) Corp.		8. IF INCORPORATED, GIVE STATE OF INCORPORATION North Dakota		9. DATE OF INCORPORATION May 1, 1989	
10. NAME AND ADDRESS OF OWNER REPRESENTATIVE(S) TO SERVE IN THIS APPLICATION. (First person listed will receive all papers)				F E E S R E C E I V E D FILING AND EXAMINATION FEES: \$ 4382.00 DATE 10/28/2005 CERTIFICATION FEE: \$ 768.00 DATE 10/13/2006	
Michael McMullen Dept. of Plant Sciences NDSU PO Box 5051 Fargo, ND 58105-5051 Dale Zetocha NDSU Research Foundation 1735 NDSU Research Park Drive PO Box 5002 Fargo, ND 58105-5002					
11. TELEPHONE (include area code) 701-231-8165		12. FAX (include area code) 701-231-8474		13. E-MAIL michael.mcmullen@ndsu.edu; dale.zetocha@ndsu.	
14. CROP KIND (Common Name) oats		16. FAMILY NAME (Botanical) Gramineae, Aveneae		18. DOES THE VARIETY CONTAIN ANY TRANSGENES? (OPTIONAL) <input type="checkbox"/> YES <input checked="" type="checkbox"/> NO IF SO, PLEASE GIVE THE ASSIGNED USDA-APHIS REFERENCE NUMBER FOR THE APPROVED PETITION TO DEREGULATE THE GENETICALLY MODIFIED PLANT FOR COMMERCIALIZATION.	
15. GENUS AND SPECIES NAME OF CROP Avena sativa		17. IS THE VARIETY A FIRST GENERATION HYBRID? <input type="checkbox"/> YES <input checked="" type="checkbox"/> NO		20. DOES THE OWNER SPECIFY THAT SEED OF THIS VARIETY BE SOLD AS A CLASS OF CERTIFIED SEED? (See Section 83(a) of the Plant Variety Protection Act) <input type="checkbox"/> YES (If "yes", answer items 21 and 22 below) <input checked="" type="checkbox"/> NO (If "no", go to item 23)	
19. CHECK APPROPRIATE BOX FOR EACH ATTACHMENT SUBMITTED (Follow instructions on reverse)		21. DOES THE OWNER SPECIFY THAT SEED OF THIS VARIETY BE LIMITED AS TO NUMBER OF CLASSES? <input type="checkbox"/> YES <input type="checkbox"/> NO IF YES, WHICH CLASSES? <input type="checkbox"/> FOUNDATION <input type="checkbox"/> REGISTERED <input type="checkbox"/> CERTIFIED			
a. <input checked="" type="checkbox"/> Exhibit A. Origin and Breeding History of the Variety b. <input checked="" type="checkbox"/> Exhibit B. Statement of Distinctness c. <input checked="" type="checkbox"/> Exhibit C. Objective Description of Variety d. <input checked="" type="checkbox"/> Exhibit D. Additional Description of the Variety (Optional) e. <input checked="" type="checkbox"/> Exhibit E. Statement of the Basis of the Owner's Ownership f. <input checked="" type="checkbox"/> Voucher Sample (2,500 viable untreated seeds or, for tuber propagated varieties, verification that tissue culture will be deposited and maintained in an approved public repository) \$4,382. g. <input checked="" type="checkbox"/> Filing and Examination Fee (\$3,652), made payable to "Treasurer of the United States" (Mail to the Plant Variety Protection Office)		22. DOES THE OWNER SPECIFY THAT SEED OF THIS VARIETY BE LIMITED AS TO NUMBER OF GENERATIONS? <input type="checkbox"/> YES <input type="checkbox"/> NO IF YES, SPECIFY THE NUMBER 1,2,3, etc. FOR EACH CLASS. <input type="checkbox"/> FOUNDATION <input type="checkbox"/> REGISTERED <input type="checkbox"/> CERTIFIED (If additional explanation is necessary, please use the space indicated on the reverse.)			
23. HAS THE VARIETY (INCLUDING ANY HARVESTED MATERIAL) OR A HYBRID PRODUCED FROM THIS VARIETY BEEN SOLD, DISPOSED OF, TRANSFERRED, OR USED IN THE U. S. OR OTHER COUNTRIES? <input checked="" type="checkbox"/> YES <input type="checkbox"/> NO IF YES, YOU MUST PROVIDE THE DATE OF FIRST SALE, DISPOSITION, TRANSFER, OR USE FOR EACH COUNTRY AND THE CIRCUMSTANCES. (Please use space indicated on reverse.)		24. IS THE VARIETY OR ANY COMPONENT OF THE VARIETY PROTECTED BY INTELLECTUAL PROPERTY RIGHT (PLANT BREEDER'S RIGHT OR PATENT)? <input checked="" type="checkbox"/> YES <input type="checkbox"/> NO IF YES, PLEASE GIVE COUNTRY, DATE OF FILING OR ISSUANCE AND ASSIGNED REFERENCE NUMBER. (Please use space indicated on reverse.)			
25. The owners declare that a viable sample of basic seed of the variety has been furnished with application and will be replenished upon request in accordance with such regulations as may be applicable, or for a tuber propagated variety a tissue culture will be deposited in a public repository and maintained for the duration of the certificate. The undersigned owner(s) is(are) the owner of this sexually reproduced or tuber propagated plant variety, and believe(s) that the variety is new, distinct, uniform, and stable as required in Section 42, and is entitled to protection under the provisions of Section 42 of the Plant Variety Protection Act. Owner(s) is (are) informed that false representation herein can jeopardize protection and result in penalties.					
SIGNATURE OF OWNER Dale Zetocha		SIGNATURE OF OWNER			
NAME (Please print or type) Dale Zetocha		NAME (Please print or type)			
CAPACITY OR TITLE Executive Director		DATE 10/26/05		CAPACITY OR TITLE DATE	

INSTRUCTIONS

GENERAL: To be effectively filed with the Plant Variety Protection Office (PVPO), **ALL** of the following items must be **received** in the PVPO: (1) Completed application form signed by the owner; (2) completed exhibits A, B, C, E; (3) for a seed reproduced variety at least 2,500 viable untreated seeds, for a hybrid variety at least 2,500 untreated seeds of each line necessary to **reproduce** the variety, or for tuber reproduced varieties verification that a viable (*in the sense that it will reproduce an entire plant*) tissue culture will be deposited and maintained in an approved public repository; (4) check drawn on a U.S. bank for \$3,652 (\$432 filing fee and \$3,220 examination fee), payable to "Treasurer of the United States" (See Section 97.6 of the Regulations and Rules of Practice.) Partial applications will be held in the PVPO for not more than 90 days, then returned to the applicant as unfilled. Mail application and other requirements to Plant Variety Protection Office, AMS, USDA, Room 401, NAL Building, 10301 Baltimore Avenue, Beltsville, MD 20705-2351. **Retain one copy for your files.** All items on the face of the application are self explanatory unless noted below. Corrections on the application form and exhibits must be initialed and dated. **DO NOT** use masking materials to make corrections. If a certificate is allowed, you will be requested to send a check payable to "Treasurer of the United States" in the amount of \$432 for issuance of the certificate. Certificates will be issued to owner, not licensee or agent.

Plant Variety Protection Office

Telephone: (301) 504-5518

FAX: (301) 504-5291

Homepage: <http://www.ams.usda.gov/science/pvpo/pvpindex.htm>

To avoid conflict with other variety names in use, the applicant must check the appropriate recognized authority and provide evidence that name has been cleared by the appropriate recognized authority before the Certificate of Protection is issued. For example, for agricultural and vegetable crops, contact: Seed Branch, AMS, USDA, 10301 Baltimore Avenue, Suite 401 NAL Building, Beltsville, MD 20705. Telephone: (301) 504-5682 <http://www.ams.usda.gov/lsg/seed.htm>.

ITEM

- 19a. Give: (1) the genealogy, including public and commercial varieties, lines, or clones used, and the breeding method;
(2) the details of subsequent stages of selection and multiplication;
(3) evidence of uniformity and stability; and
(4) the type and frequency of variants during reproduction and multiplication and state how these variants may be identified
- 19b. Give a summary of the variety's distinctness. Clearly state how this application variety may be distinguished from all other varieties in the same crop. If the new variety is most similar to one variety or a group of related varieties:
- (1) identify these varieties and state all differences objectively;
 - (2) attach statistical data for characters expressed numerically and demonstrate that these are clear differences; and
 - (3) submit, if helpful, seed and plant specimens or photographs (prints) of seed and plant comparisons which clearly indicate distinctness.
- 19c. Exhibit C forms are available from the PVPO Office for most crops; specify crop kind. Fill in Exhibit C (Objective Description of Variety) form as completely as possible to describe your variety.
- 19d. Optional additional characteristics and/or photographs. Describe any additional characteristics that cannot be accurately conveyed in Exhibit C. Use comparative varieties as is necessary to reveal more accurately the characteristics that are difficult to describe, such as plant habit, plant color, disease resistance, etc.
- 19e. Section 52(5) of the Act requires applicants to furnish a statement of the basis of the applicant's ownership. An Exhibit E form is available from the PVPO.
20. If "Yes" is specified (*seed of this variety be sold by variety name only, as a class of certified seed*), the applicant **MAY NOT** reverse this affirmative decision after the variety has been sold and so labeled, the decision published, or the certificate issued. However, if "No" has been specified, the applicant may change the choice. (See Regulations and Rules of Practice, Section 97.103).
23. See Sections 41, 42, and 43 of the Act and Section 97.5 of the regulations for eligibility requirements.
24. See Section 55 of the Act for instructions on claiming the benefit of an earlier filing date.

22. CONTINUED FROM FRONT (Please provide a statement as to the limitation and sequence of generations that may be certified.)

23. CONTINUED FROM FRONT (Please provide the date of first sale, disposition, transfer, or use for each country and the circumstances, if the variety (including any harvested material) or a hybrid produced from this variety has been sold, disposed of, transferred, or used in the U.S. or other countries.)

Stark was distributed to the North Dakota Crop Improvement Association under contract for seed increase. The first certified seed tag to a crop improvement grower was issued November 1, 2004.

24. CONTINUED FROM FRONT (Please give the country, date of filing or issuance, and assigned reference number, if the variety or any component of the variety is protected by intellectual property right (Plant Breeder's Right or Patent).)

'Paul' oat variety was issued under PVP Certificate No. 9600002 issued March 13, 1996.

NOTES: It is the responsibility of the applicant/owner to keep the PVPO informed of any changes of address or change of ownership or assignment or owner's representative during the life of the application/certificate. The fees for filing a change of address; owner's representative; ownership or assignment; or any modification of owner's name is specified in Section 97.175 of the regulations. (See Section 101 of the Act, and Sections 97.130, 97.131, 97.175(h) of the Regulations and Rules of Practice.)

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0581-0055. The time required to complete this information collection is estimated to average 1.4 hours per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information.

The U.S. Department of Agriculture (USDA) prohibits discrimination in all its programs and activities on the basis of race, color, national origin, gender, religion, age, disability, sexual orientation, marital or family status, political beliefs, parental status, or protected genetic information. (Not all prohibited bases apply to all programs.) Persons with disabilities who require alternative means for communication of program information (Braille, large print, audiotape, etc.) should contact USDA's TARGET Center at 202-720-2600 (voice and TDD).

To file a complaint of discrimination, write USDA, Director, Office of Civil Rights, Room 326-W, Whitten Building, 14th and Independence Avenue, SW, Washington, DC 20250-9410 or call 202-720-5964 (voice and TDD). USDA is an equal opportunity provider and employer.

19 a. Exhibit A. Origin and Breeding History of 'Stark'

Pedigree

ND900677/'Paul'

ND900677 = ND863437/ND863384

ND863437 = W80-19/SO81136

W80-19 = Germplasm line with unknown parentage
received from Agriculture and Agri-food Canada,Winnipeg that possessed crown rust resistance genes Pc-55
and Pc-56.

SO81136 = Otana/Cascade

ND863384 = SD800043/W80-19

SD800043 = 'Noble'/'Dal'/'Nodaway 70'

Experimental Designation ND960736

19 a. Exhibit A. Origin and Breeding History of 'Stark'

Breeding Method –

Modified single seed descent and pedigree method

Selection and Multiplication –	Stage of development	Selection Criteria
1992 Fall greenhouse	Final cross	
1993 Spring greenhouse	F ₁	F ₁ plants were uniform and seed from 3 plants was bulked to produce F ₂ population
1993 Field	F ₂ selection of single panicle	F ₂ population was segregating for crown rust and stem rust resistance in the field. Plants exhibiting multiflorous naked seeded phenotype and resistant to both stem rust and stem rust were selected for advancement.
1993 Fall greenhouse	F ₃ single seed descent accompanied by screening for seedling resistance to critical races of stem and crown rust.	Seedlings were inoculated with composite of crown rust races that were virulent on Pc-38 and Pc-39 and with stem rust race NA27. Seedlings exhibiting a resistant infection type were grown to maturity and seed from individual resistant F ₃ plants that exhibited the naked seed characteristic were advanced to the field.
1994 Field	F ₄ planted in hill plots from seed of single F _{3,4} panicle F ₄ panicles harvested from selected hill plots	Panicles from plants in hill plots exhibiting the naked seed characteristic, stem rust and crown rust resistance, along with resistance to lodging and tolerance to barley yellow dwarf virus were harvested to provide seed for advancement to the F ₅ .

19 a. Exhibit A. Origin and Breeding History of 'Stark'

Breeding Method –

Modified single seed descent and pedigree method

Selection and Multiplication –	Stage of development	Selection Criteria
1995 Field	Seed from F ₄ panicles were planted to produce paired hill plots. A selected paired hill plot was harvested in bulk to produce an F _{4.5} breeding line that was subsequently designated ND960736 that became the source of Stark breeder's seed.	Hill plots exhibiting homogeneity of crown rust resistance and stem rust resistance were selected for harvest. Lodging resistance, expression of the naked seed characteristic, and visual selection of kernel morphology were considered to further select plots that were identified for harvest. Harvested lines were evaluated as seedlings in the greenhouse using stem rust race NA27 and a composite of crown rust races to identify lines homogeneous for resistance to these diseases. These selected lines were advanced to the F ₆ generation.
1996 Field	F ₆ Preliminary screening trial – Fargo location, one replication with repeating checks.	Selection was based on lodging resistance, medium heading date, high grain yield, high test weight, expression of naked seed characteristic, and resistance to stem and crown rust in the field. Stem rust and crown rust seedling resistance evaluation was repeated in the greenhouse to identify homogeneous resistant lines.
1997 Field	F ₇ Naked Preliminary Yield Trial (NPYT) – Two ND locations, two replications per location ND960736 assigned to line.	Selection was based on lodging resistance, medium heading date, high grain yield, high test weight, expression of naked seed characteristic, and resistance to stem and crown rust in the field. Seedling stem and crown rust.

19 a. Exhibit A. Origin and Breeding History of 'Stark'

Breeding Method –

Modified single seed descent and pedigree method

Selection and Multiplication –	Stage of development	Selection Criteria
1998 Field	F ₈ Coop Naked Oat Trial (CNOT), 2 ND locations with 3 replications, 8 locations in other states and provinces of Canada. NPYT, 2 ND locations, 2 replications Increase plot rouged of non-naked variants to initiate production of breeder seed.	Evaluation was based on lodging resistance, medium heading date, high grain yield, high test weight, naked seed, and resistance to stem and crown rust in the field. Stem rust and crown rust seedling resistance was evaluated in the greenhouse. Stem rust and crown rust seedling resistance evaluation was repeated in the greenhouse.
1999 Field	F ₉ North Dakota Oat Variety Trials at ten locations (NDOVT) and CNOT at 9 locations. Increase plot evaluated for homogeneity and non-naked variants were removed.	ND960736 that became Stark was determined to produce higher grain yield and lower groat protein concentration than Paul, medium high test weight, and more than 99% naked seed. Stem rust and crown rust resistance evaluation at many locations indicated ND960736 had stable resistance to stem rust races NA27 and NA67. Stem rust and crown rust seedling resistance evaluation was repeated in the greenhouse.

19 a. Exhibit A. Origin and Breeding History of 'Stark'

Breeding Method –

Modified single seed descent and pedigree method

Selection and Multiplication –	Stage of development	Selection Criteria
2000 ND Field	F ₁₀ NDOVT at 10 locations and CNOT at 8 locations Increase and purification in drill strip at Fargo	ND960736 that became Stark was determined to produce high grain yield, medium high test weight, and More than 99% naked seed Stem rust and crown rust resistance was evaluated at many locations and ND960736 was identified to have stable crown rust resistance and resistance to stem rust races NA27 and NA67.
2001 Field	F ₁₁ NDOVT at 10 locations, CNOT at 8 locations. Increase and purification in drill strip at Fargo.	Evaluation continued for all characteristics evaluated in 2000
2002 Field	F ₁₂ NDOVT Preliminary large increase by Foundation Seed Stocks form F ₁₁ Breeder's Seed	Evaluation continued for all characteristics evaluated in 2000
2003 Field	F ₁₃ NDOVT 8 locations Foundation seed increase and release as Stark	Evaluation continued for all characteristics evaluated in 2000

19 a. Exhibit A. Origin and Breeding History of 'Stark'

Evidence of uniformity and stability:

Stark has been observed to be uniform and stable for stem rust resistance and crown rust resistance for eight generations from the original $F_{4.5}$ that was designated ND60736 in 1996 until release in 2003. Stark has been observed to produce up to 1% variants that are non-naked and non-multiflorous. The frequency of these variants has not changed for eight generations since they were observed in the F_8 generation in 1998. Stark appears otherwise uniform and stable.

The type and frequency of variants during reproduction and multiplication and how these variants may be identified:

The non-naked seed variants comprise less than 1% of the Stark plants. The non-naked variants are conspicuous because the spikelets are not multiflorous as is typical of naked oats.

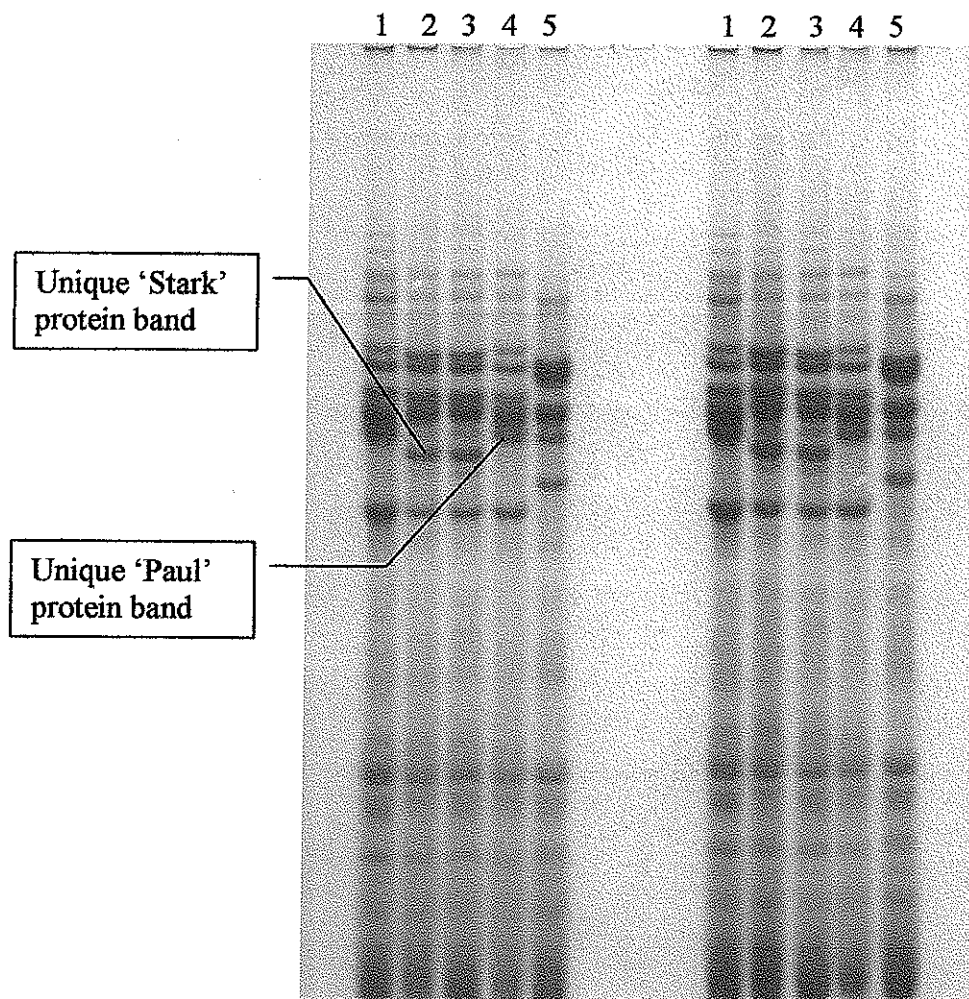
19b. Exhibit B. Statement of distinctness (revised).

'Stark' is a spring oat that is most similar to 'Paul' in appearance. Paul and Stark are the only naked oat cultivars that possess the stem rust resistance gene complex *pg-a* that confers resistance to stem rust race NA67. Evidence for the presence of *pg-a* is provided by the highly resistant (;) infection type produced on seedlings in the greenhouse after inoculation with stem rust race NA67 (Table 6). Stark can be distinguished from Paul by acid polyacrylamide gel electrophoresis (PAGE) analysis of seed protein as illustrated in the attached figure. The PAGE procedure for oats was modified by the North Dakota State Seed Department from the ISTA (International Seed Testing Association) procedure for variety testing of wheat, with a modification on the gel run time. The ISTA procedure is referenced in the 1992 Handbook of Variety Testing edited by R. J. Cooke. This is an International Seed Testing Association Publication. The procedure is on pages 2-5 through 2-6. A copy of the relevant section of the handbook (pages 2-5 and 2-6) is attached. The modified procedure for oat seed protein analysis used by the North Dakota State Seed Department also is attached. Application of the PAGE procedure to Paul and Stark provides a clear distinction in protein banding pattern to distinguish between the two cultivars.

200600022

9/26/06

Oat Seed Protein Electrophoresis Test Results



NDSSD Acid PAGE Analysis of Oat seed protein. Samples submitted for testing on 9-20-06 by NDSU Plant Science Department (Dr. McMullen). Sample lanes represent: 1 = Paul Oat control; 2 = Stark Oat control; 3 = L2601132 (Stark sample); 4 = L2601133 (Paul sample); and 5 = Morton Oat control.

A. ISTA Standard Reference Method for the Identification of Varieties of Wheat and Barley by Polyacrylamide Gel Electrophoresis (PAGE)

A. 1. Principle

The alcohol-soluble proteins (gliadins from wheat, hordeins from barley) are extracted from seeds and separated by PAGE at pH 3.2. The pattern of protein bands produced (electrophoregram) is related to genetic constitution and can be considered as a 'fingerprint' of a variety. The 'fingerprints' can be used to identify unknown samples and mixtures, by single seed analysis.

As a guideline, it is recommended that 100 seeds are used. Very precise estimates of varietal purity may require a larger sample. If a comparison is being made with a standard value, sequential testing using batches of 50 seeds can be undertaken in order to minimise the workload. A simple check on the identity of a single major constituent of a seed lot can be done using less than 50 seeds.

A. 2. Apparatus and Equipment

A. 2.1 The Pharmacia GE-2/4 electrophoresis apparatus and EPS 400/500 power supply have been successfully used, but any suitable vertical electrophoresis system eg. Desaga, BioRad, Biometra should give comparable results.

A. 2.2 Chemicals

All chemicals should be of 'Analytical Reagent' grade or better.

Acrylamide ('specially purified for electrophoresis')
Bisacrylamide ('specially purified for electrophoresis')
Urea
Glacial acetic acid
Glycine
Ferrous sulphate
Ascorbic acid
Hydrogen peroxide (or ammonium persulphate and TEMED)
Monothioglycerol (or 2-mercaptoethanol)
Pyronin G (or methyl green)
Trichloroacetic acid
Ethanol
2-chloroethanol
PAGE Blue G-90 (or PAGE Blue 83) (or any reagent equivalent to the 'Coomassie Brilliant Blue' G or R series of dyes).

A. 2.3 Solutions

A. 2.3.1 Extraction solution

Wheat: Pyronin G (or methyl green) 0.05%
2-chloroethanol 25%

Keep cold.

Barley: Pyronin G (or methyl green) 0.05%
2-chloroethanol 20%
containing Urea 18%
monothioglycerol 1%
(or 2-mercaptoethanol) 1%

Keep cold or prepare fresh.

A. 2.3.2 Tank buffer solution:

Glacial acetic acid 4 ml
Glycine 0.4 g

Made up to 1l with water; keep cold.

A. 2.3.3 Gel buffer solution:

Glacial acetic acid 20 ml
Glycine 1.0 g

Made up to 1l with water; keep cold.

A. 2.3.4 Staining solution:

1 Trichloroacetic acid 100 g
Water 1 l
2 PAGE Blue G-9 (or PAGE Blue 83) 1 g
Ethanol 100 ml

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A. 3. Procedure

A. 3.1 Protein extraction

Single seeds are crushed with pliers or a similar implement and transferred to 1.5 ml polypropylene centrifuge tubes or to the wells of a micro-titer plate. Extraction solution (A. 2.3.1) (0.2 ml for wheat, 0.3 ml for barley) is added, the contents of the tubes or plates are thoroughly mixed and the tubes are allowed to stand (covered or sealed) overnight at room temperature. The tubes are centrifuged at 18000 xg and the supernatants used for electrophoresis.

A. 3.2 Preparation of the gel

Clean and dry gel cassettes are assembled, according to the design of the equipment. Treating the glass plates with silicon prior to assembly can facilitate subsequent removal of the gel. The gel cassettes can incorporate a plastic backing sheet (eg 'Gel Bond PAG', FMC Corporation). This supports the gel during subsequent operations.

Gel Medium

Gel buffer (A.2.3.3)	60 ml <i>ca</i>
Acrylamide	10 g
Bisacrylamide	0.4 g
Urea	6 g
Ascorbic acid	0.1 g
Ferrous sulphate	0.005 g

Stir the solution and make up to 100 ml with

Stock gel buffer (A.2.3.3.)

Add, mixing quickly

*freshly prepared 0.6% (v/v) Hydrogen peroxide 0.35 ml
per 100 ml gel medium*

Pour the gel.

(Note: the gel mixture can be cooled to near freezing prior to the addition of the peroxide.)

Polymerisation should be complete in 5–10 minutes. If not, it may be necessary to adjust the concentration of hydrogen peroxide added. An acrylic 'comb' is placed in the top of the cassette, to make wells in the gel. The gel mixture should over-fill the cassette, or be over-layered with water, to ensure satisfactory polymerisation of the upper surface.

Note that as an alternative to the hydrogen peroxide catalyst, it is possible to use ammonium persulphate (0.1 ml of 10% solution, freshly prepared) and TEMED (0.3 ml) added to the gel mixture prior to pouring the gel.

A. 3.3 Electrophoresis

The acrylic comb is removed from the gel and the sample wells washed with tank buffer (A.2.3.2). The tank is filled with an appropriate volume of buffer (A. 2.3.2) (depending on the equipment used). Samples (10–20 µl) are loaded into the wells and the gel placed in the tank, ensuring that the sample wells are completely filled. Electrophoresis is carried out at no more than 500 V (constant voltage) for twice the time taken for the pyronin G marker dye to leave the gel, or three times if methyl green is used as a tracking dye. It must be remembered that the anode (positive electrode) is at the origin (top of the gel) in this system and the polarity of the electric field should be adjusted accordingly. Water should be circulated through the buffer tank to maintain the temperature at 15–20° C.

A. 3.4 Fixing and staining

The gel cassette is removed from the tank, opened and the gel placed in a plastic or glass box containing 5–10 ml of 1% PAGE Blue G90 (or PAGE Blue 83) in 200 ml of 10% trichloroacetic acid (A.2.3.4). Staining is complete in 1–2 days at room temperature and de-staining is not usually needed. Precipitated stain should be scraped from the surface of the gel. The gel is washed in water to enhance the stain and can then be examined or photographed. Any blue background in the gel is removed by washing in 10% trichloroacetic acid. Gels can be stored in polythene bags at 4° C for many months without deterioration.

Typical results produced using the above procedure, and methods of utilising and reporting the electrophoretic data are presented in Section 3 of the Handbook.

One of the benefits of the ISTA standard reference method is that it can be utilised, with little or no modification, for prolamin analysis and variety identification in other cereals such as oats, durum wheat, triticale and rye (4). The Electrophoresis Working Group will be organising a collaborative test of the method for oats identification, with a view to including this in the International Rules. Rice and maize varieties have also been reported as being successfully analysed using essentially this procedure.

**ISTA Varietal Identification Procedure for Wheat and Oats Using
Bulked Seed Analysis (North Dakota State Seed Department Protocol-
revised 2006)**

1. **Sample preparation:** Place approximately 100 seed into a coffee grinder and grind for about 20 seconds. Pass sample through a #9 and #4 dodder/purity test sieves (Hoffman Manufacturing, Inc.). Weigh out 0.15g of ground and sieved sample and place into a 1.8ml capped centrifuge tube containing 0.6ml of 2CE extraction buffer (see recipe below). Vortex and incubate overnight at 4°C. Prior to use, centrifuge at 8000g for 5 minutes. Apply 6-8ul of supernatant to the gel. Prepare control samples in the same manner as the samples.
2. **Gel preparation:** Prepare a 10% acrylamide gel using the reagents and procedures listed below. The gel should be prepared a day in advance or 2 hours prior to usage. Glass plates are cleaned and coated with Rain-X to aid in gel removal from the glass. Our lab uses the Bio-Rad Protein II system with 16x20cm gels and a 25 well comb. Electrophoresis is run using a cooling system to maintain a gel temperature of 20°C.
3. **Electrophoresis:** Electrophoresis is carried out under constant voltage of 500 volts for 5.5 hours for wheat and 2.5 hours for oats.
4. **Gel staining:** After electrophoresis, gels are removed and placed into trays containing 300 ml distilled water with 50 g trichloroacetic acid and 10ml of 2% coomassie blue G-250 in 95% EtOH (see below). Stain gels overnight for best results. Destain gels as necessary with water to remove excess stain. Gels can be scanned or dried using cellophane sheets to keep permanently.
5. **Gel interpretation:** Gels are scored visually using a light box. Interpretation can also be conducted using our Kodak Imaging System.
6. **Recipes:**

2CE extraction buffer

150ml 2-chloroethanol
350ml DI water
1 ml of 1% methyl green
store at 4°C

Gel Buffer 1X

20 ml glacial acetic acid
1.0g glycine
1.45g ascorbic acid
add DI water to 1 liter
adjust pH to 3.1 if necessary

10X Tank Buffer

160ml glacial acetic acid
16g glycine
DI water to 4 liters

Gel recipe (2 gels)

68.75ml gel buffer
31.25ml 40% acrylamide
25ml 2% bis-acrylamide
300 ul of 0.1% FeSO₄
use 110 ul of H₂O₂ to polymerize

G-250 stain solution

4 g G-250
200 ml 95% ethanol

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**U.S. DEPARTMENT OF AGRICULTURE
AGRICULTURAL MARKETING SERVICE
SCIENCE AND TECHNOLOGY
PLANT VARIETY PROTECTION OFFICE
BELTSVILLE, MD 20705**

Exhibit C

**OBJECTIVE DESCRIPTION OF VARIETY
Oat (*Avena* spp.)**

NAME OF APPLICANT (S) Michael S. McMullen	TEMPORARY OR EXPERIMENTAL DESIGNATION ND960736	VARIETY NAME 'Stark'
ADDRESS (Street and No. or RD No., City, State, Zip Code, and Country) Dept. of Plant Sciences, NDSU, PO Box 5051 Fargo, ND 58105-5051		FOR OFFICIAL USE ONLY PVPO NUMBER 200600022

Place the appropriate number that describes the varietal character of this variety in the boxes below. Place a zero in the first box (i.e. or) when the number is either 99 or less or 9 or less.

1. SPECIES:

1 = *Sativa* 2 = *Byzantina* 3 = Other (Specify) _____

2. GROWTH HABIT:

1 = Winter 2 = Semi-Winter 3 = Spring
 Juvenile Growth: 1 = Prostrate 2 = Semi-Prostrate 3 = Erect

3. MATURITY: (50% Flowering)

Number of days
 No. Days Earlier Than * _____
 Same as Check * Paul
 No. of Days Later Than * _____
 5 Season: 1 = Very Early (Jaycee) 2 = Early (Nodaway 70) 3 = Midseason (Clintford)
 4 = Late (Lodi) 5 = Very Late (Gerry) 6 = Extremely Late (Mackinaw)

4. PLANT HEIGHT: (From Soil Level to Top of Head)

cm Tall
 cm Shorter Than * _____
 Same as Check * _____
 cm Taller Than * Paul

* Relative to a Commercial Variety Grown in the Same Trial

5. STEM:

Diameter: 1 = Fine (Kherson) 2 = Medium (Clintford) 3 = Coarse (Nodaway 70)
 Hairiness at Upper Culm Nodes: 1 = Hairless 2 = Hairy
 Mature Stem Color 1 = Yellow 2 = Reddish

6. LEAF: (Leaf Color: The Royal Horticultural Society's or any recognized color chart should be used to determine the leaf color of the described variety.)

Carriage: 1 = Drooping (Random) 2 = Erect (Walken)
 Color: 1 = Yellow Green 2 = Light Green 3 = Dark Green 4 = Blue Green
 mm Width (First leaf below flag leaf) Leaf Margin: 1 = Glabrous 2 = Ciliate
 Ligule: 1 = Absent 2 = Present Leaf Sheath: 1 = Hairless 2 = Hairy

7. HEAD:

Panicle Shape: 1 = Equilateral 2 = Intermediate 3 = Side Panicle (Unilateral)
 Attachment of Lower Whorl of Branches: 1 = First Node 2 = Second Node (False Node)
 Panicle Size: 1 = Small (Yancey) 2 = Medium (Walken) 3 = Large (Markton)
 Panicle Width: 1 = Narrow (Gopher) 2 = Midbroad (Yancy) 3 = Broad (Nodaway 70)
 cm Panicle Length Number of Branches Number of Whorls of Branches
 Position of Branches: 1 = Ascending (Yancey) 2 = Spreading (Cayuse) 3 = Drooping (Markton)
4 = Pectinate (White Tarter) 5 = Confused (Storm King)

8. RACHIS:

1 = Recurved (Yancey) 2 = Erect (Walken) mm Second Floret Rachilla Segment Length
 Second Floret Rachilla Segment: 1 = Hairless 2 = Hairy Rachilla Hairs: 1 = Short 2 = Long

9. SPIKELET:

Spikelet Separation by: 1 = Abscission 2 = Semi-Abscission 3 = Fracture
 Floret Separation by: 1 = Disarticulation 2 = Heterofracture 3 = Basifracture
 Florets per Spikelet (Mean no.)

10. GLUMES: (Glume Color: The Royal Horticultural Society's or any recognized color chart should be used to determine the leaf color of the described variety.)

mm Width mm Length No. of Veins on Glumes Color: 1 = White 2 = Yellow 3 = Red 4 = Striped

11. LEMMA: (Lemma Color: The Royal Horticultural Society's or any recognized color chart should be used to determine the leaf color of the described variety.)

mm Length Color: 1 = White 2 = Yellow 3 = Red 4 = Gray 5 = Black
 Hairiness of Dorsal Surface: 1 = Hairless 2 = Hairy

12. AWN: (First Floret)

Occurrence: 1 = Absent (Walken) 2 = Infrequent (Yancey) 3 = Common (Chilocco) 4 = Frequent (Random)
 Type: 1 = Non-twisted 2 = Twisted 3 = Twisted Geniculate
 mm Awn Length

13. SEED:

☒ 2
☒ 1

Florescence Under Ultraviolet Light:

1 = Florescent

2 = Non-florescent

Basal Hair:

1 = Absent (Florida 501)

4 = Several to Numerous (Florilee)

2 = Absent to Few (Yancey)

5 = Numerous (Red Rustproof)

3 = Few to Several (Lee)

☐ . ☐

mm Basal Hair Length

☐ ☐ . ☐

gms per 1000 Seeds

☐ 2 ☐ 5

mg Groat Weight (Each)

☐ ☐ . ☐

% Groat Protein

☐ ☐ . ☐

% Groat Oil

14. INSECTS: (0 = Not Tested 1 = Susceptible 2 = Resistant)

☐ 0

Cereal Leaf Beetle

☐ 0

Bluegrass Billbug

☐ 0

Grain Bug (C. Sayi)

☐ 0

Nematode (Type)

☐ 0

Green Bug (Biotype)

☐

Other (Specify)

15. DISEASE: (0 = Not Tested 1 = Susceptible 2 = Resistant)

☐

Halo Blight

☐

Powdery Mildew

☐

Septoria Leaf Blotch

☐

Soil-Borne Mosaic

☐
Helminthosporium
Leaf Blotch
☐

Yellow Dwarf Virus

☐

Victoria Blight

☐

Other (Specify)

☐

Specify Races Tested:

☐

Crown Rust

☐

Stem Rust

☐

Covered Smut

☐

Loose Smut

Races Susceptible	Races Resistant
	CR13, CR36, CR152, CR169, CR181
	NA26, NA27, NA67
	Not Identified

16. INDICATE THE VARIETY YOU BELIEVE MOST CLOSELY TO RESEMBLE THAT SUBMITTED:

CHARACTER	VARIETY	CHARACTER	VARIETY
Plant Tilling	Paul	Leaf Color	Paul
Leaf Size	Paul	Leaf Carriage	Paul
Seed Color	Paul	Seed Shape	Paul

COMMENTS:

Stark produces multiflorous spikelets and naked seed under normal threshing conditions.

19d. Exhibit D. Additional Description of the Variety

Stark has been evaluated in replicated trials in North Dakota since 1997. During three years of evaluation in North Dakota Oat Variety trials that include 33 location/years, it has exhibited grain yield potential greater than 'Paul' (Tables 1, 2, and 3) and groat yields that are equivalent to the highest yielding conventional lines. Stark is relatively late maturing, but heads slightly earlier than Paul (Table 1). Although Stark is relatively tall, it has very good straw strength and lodging resistance (Table 1).

The characteristics of Stark are similar to 'Paul', but in North Dakota trials, Stark produced approximately a 10% yield advantage relative to Paul (Table 1). The grain oil concentration and grain protein concentration of Stark are not as high as Paul (Tables 1 and 5). Stark, like Paul, is susceptible to some races of crown rust (incited by *Puccinia coronata* cda. f. sp. *avenae* Eriks) that occur in some areas of North Dakota (Table 7). Stark expresses effective resistance to all races of stem rust (incited by *Puccinia graminis avenae*) in North Dakota, including NA67 that is virulent on nearly all other oat cultivars (Table 6). Stark has good barley yellow dwarf virus tolerance and strong straw.

In North Dakota, Stark produces high grain yield and under normal threshing conditions produces more than 95% naked groats. Characteristic of naked oats, spikelets of Stark are multiflorous with elongated rachilla relative to conventional oats. The lemma and palea do not adhere to the groat. Approximately 1% of plants do not produce multiflorous spikelets and the groats do not thresh free from the hulls in these plants. Culms and leaf margins of Stark are glabrous and ligules are present. Stark has equilateral panicles with ascending branches with a tendency toward unilateral shape at heading. Awns are absent.

19d. Exhibit D. Additional Description of the Variety

Table 1.

Stark compared to selected lines in 2000-2003 North Dakota Oat Variety Trial 2000-2003.

Genotype	Grain Yield	Test Wt.	% Groat	Thin Kernels <5/64"	Whole Oat Protein	Head >May 31	Plant Height	Lodge Score	Kernel Wt.	Groat Lipid
	bu/a	lb/bu	%	Proportion	%	Days	cm	0-5	mg	%
AC Assiniboia	118.7	35.4	75.0	0.08	14.9	32.3	103	1.2	38.5	8.8
BUFF	--	--	--	--	--	24.1			24.5	
Ebeltoft	122.3	34.6	73.4	0.11	14.3	33.2	96	1.8	37.0	8.3
HiFi	120.9	36.5	73.6	0.18	14.3	30.5	106	1.7	33.8	8.6
Hytest	95.7	39.4	76.2	0.08	18.0	26.6	111	2.8	35.3	7.8
Jerry	109.4	37.4	73.6	0.11	15.5	27.0	108	1.2	34.3	7.4
AC Kaufman	--	--	--	--	--	30.9			40.3	
Killdeer	126.3	35.2	74.2	0.11	13.2	29.3	95	2.4	33.8	7.8
AC Medallion	115.9	35.5	75.2	0.10	13.9	31.8	107	3.0	36.8	9.3
Morton	118.6	37.4	73.1	0.10	15.3	29.9	114	0.9	33.8	7.3
Otana	107.3	33.8	70.7	0.19	13.6	31.4	110	3.1	30.0	8.0
Paul	86.0	41.8	91.0	0.56	20.3	32.3	108	2.2	23.5	11.0
Youngs	117.5	34.8	74.7	0.08	15.3	31.3	113	2.4	39.0	8.5
Beach	122.0	38.0	75.3	0.10	14.4	29.6	111	0.9	34.8	9.6
Stark	94.4	41.3	91.8	0.45	18.1	31.7	110	1.8	25.3	9.3
Loc. Yrs.	33	33	32	31	30	6	4	4	4	4

Table 2

North Dakota Oat Variety Trial 2000-2003 grain yield and test weight summary.

Genotype	Grain Yield				Test Weight			
	2003 9 Loc. Mean	2002-03 2 yr. Mean	2001-03 3 yr. Mean	2000-03 4 yr. Mean	2003 9 Loc. Mean	2002-03 2 yr. Mean	2001-03 3 yr. Mean	2000-03 4 yr. Mean
	Bushels/Acre				Lb/Bushel			
AC Assiniboia	126	105.5	114.2	118.7	36.2	35.0	34.5	35.4
BUFF	92	--	--	--	45.3	--	--	--
Ebeltoft	134	110.7	118.1	122.3	35.3	34.4	33.8	34.6
HiFi	132	108.8	116.9	120.9	37.1	35.9	35.8	36.5
Hytest	109	91.0	95.9	95.7	40.4	39.4	39.0	39.4
Jerry	126	103.7	109.1	109.4	39.0	37.8	37.2	37.4
Kaufman AC	127	--	--	--	37.0	--	--	--
Killdeer	145	118.0	123.0	126.3	36.2	35.2	34.5	35.2
AC Medallion	127	102.2	112.4	115.9	36.7	35.6	35.1	35.5
Morton	131	107.0	117.6	118.6	37.9	37.0	36.9	37.4
Otana	124	104.2	107.4	107.3	35.2	34.6	33.6	33.8
Paul	96	77.1	83.3	86.0	43.1	41.8	41.1	41.8
Pinnacle AC	135	--	--	--	35.6	--	--	--
Reeves	97	--	--	--	39.2	--	--	--
Ronald AC	129	--	--	--	38.3	--	--	--
Sesqui	133	111.6	--	--	37.5	36.4	--	--
Youngs	136	110.2	116.0	117.5	35.9	34.7	33.9	34.8
Beach	135	111.2	118.5	122.0	39.2	38.0	37.4	38.0
Stark	103	85.3	89.4	94.4	39.9	40.4	39.9	41.3
Loc. Yrs.	10	16	24.0	33	9.0	14.0	24.0	34.0

19d. Exhibit D. Additional Description of the Variety

Table 3

North Dakota Oat Variety Trial 2000-2003 thin oat proportion summary.

Genotype	2000 9 Loc. Mean	2001 6 Loc. Mean	2002 8 Loc. Mean	2003 8 Loc. Mean	2002-03 2 yr. Mean	2001-03 3 yr. Mean	2000-03 4 yr. mean
----- Proportion through 5/64" sieve -----							
AC Assiniboia	0.04	0.08	0.12	0.07	0.10	0.09	0.08
Buff	--	--	--	0.67	--	--	--
Ebeltoft	0.07	0.07	0.17	0.13	0.15	0.13	0.11
HiFi	0.11	0.17	0.27	0.18	0.23	0.21	0.18
Hytest	0.05	0.07	0.13	0.07	0.10	0.09	0.08
Jerry	0.08	0.10	0.16	0.09	0.13	0.12	0.11
Kaufman	--	--	--	0.05	--	--	--
Killdeer	0.07	0.11	0.15	0.10	0.13	0.12	0.11
AC Medallion	0.06	0.09	0.16	0.11	0.13	0.12	0.10
Morton	0.06	0.07	0.17	0.12	0.13	0.12	0.10
Otana	0.14	0.16	0.25	0.20	0.21	0.21	0.19
Paul ^a	0.44	0.58	0.62	0.63	0.60	0.61	0.56
Pinnacle	--	--	--	0.07	--	--	--
Reeves	--	--	--	0.11	--	--	--
Ronald	--	--	--	0.16	--	--	--
Sesqui	--	--	0.42	0.25	0.33	--	--
Youngs	0.05	0.08	0.11	0.09	0.10	0.10	0.08
Beach	0.06	0.09	0.14	0.11	0.12	0.11	0.10
Stark	0.36	0.47	0.51	0.48	0.50	0.49	0.45
Loc. Yrs.	9	6	8	8	16	22	31

^a Excludes data from Williston 2002

Table 4

North Dakota Oat Variety Trial 2000-2003 Groat Percentage Summary.

Genotype	2000 9 Loc. Mean	2001 6 Loc. Mean	2002 8 Loc. Mean	2003 9 Loc. Mean	2002-03 2 yr. Mean	2001-03 3 yr. Mean	2000-03 4 yr. mean
----- Groat Percentage -----							
AC Assiniboia	76.7	76.2	77.8	70.0	73.7	74.3	75.0
Buff	--	--	--	95.0	--	--	--
Ebeltoft	74.3	70.8	74.5	73.3	73.9	73.1	73.4
HiFi	74.7	72.7	73.6	73.0	73.3	73.1	73.6
Hytest	77.0	74.8	76.5	75.9	76.2	75.8	76.2
Jerry	75.3	70.2	73.9	73.7	73.8	72.9	73.6
Kaufman	--	--	--	77.6	--	--	--
Killdeer	76.0	71.3	74.5	74.0	74.2	73.5	74.2
AC Medallion	75.7	74.3	76.0	74.8	75.3	75.1	75.2
Morton	73.0	73.3	73.6	72.6	73.1	73.1	73.1
Otana	74.0	67.2	71.6	68.9	70.2	69.4	70.7
Paul	89.0	89.5	91.3	93.8	92.6	91.8	91.0
Pinnacle	--	--	--	74.6	--	--	--
Reeves	--	--	--	74.8	--	--	--
Ronald	--	--	--	75.7	--	--	--
Sesqui	--	--	70.8	70.7	70.8	--	--
Youngs	74.3	73.2	75.9	74.9	75.4	74.8	74.7
Beach	75.3	75.3	76.1	74.5	75.2	75.3	75.3
Stark	93.0	90.2	93.1	90.7	91.8	91.4	91.8
Loc. Yrs.	9	6	8	9	17	23	32

19d. Exhibit D. Additional Description of the Variety

Table 5

2000-2003 ND Oat Variety Trial Whole Oat Protein Summary

Genotype	2000 7 Loc Mean	2001 6 Loc Mean	2002 8 Loc. Mean	2003 9 Loc Mean	2002-03 2 yr. Mean	2001-03 3 yr. Mean	2000-03 4 yr. mean
	%						
Assiniboia	14.5	14.3	14.7	15.9	15.3	15.1	14.9
Buff	--	--	--	18.0	--	--	--
Ebeltoft	14.6	13.3	14.7	14.3	14.4	14.1	14.3
HiFi	13.8	14.0	14.4	14.8	14.6	14.5	14.3
Hytest	17.9	17.7	18.3	17.9	18.1	18.0	18.0
Jerry	15.7	14.5	15.8	15.9	15.8	15.5	15.5
AC Kaufman	--	--	--	13.3	--	--	--
Killdeer	13.8	12.0	13.3	13.5	13.4	13.0	13.2
AC Medallion	14.0	12.9	14.5	14.2	14.3	13.9	13.9
Morton	15.5	14.9	15.4	15.4	15.4	15.3	15.3
OTANA	13.8	12.9	13.7	13.9	13.8	13.6	13.6
PAUL	21.3	18.5	21.3	19.9	20.6	20.0	20.3
AC Pinnacle	--	--	--	12.9	--	--	--
Reeves	--	--	--	17.1	--	--	--
AC Ronald	--	--	--	15.0	--	--	--
Sesqui	--	--	17.0	17.4	17.2	--	--
Youngs	15.2	14.4	15.9	15.4	15.6	15.3	15.3
Beach	14.5	13.6	14.8	14.5	14.7	14.4	14.4
Stark	19.3	16.8	19.9	16.5	18.1	17.8	18.1
Loc. Yrs.	7	6	8	9	17	23	30

Table 6

2000-2003 Oat stem rust evaluation.

	Fargo Field 2002			2003	Green House Seedling Evaluation				
Genotype	Yield Plot		Hill Plot	Hill Plot	2000	2001	2002		2003
	Rep 1	Rep 2			NA67	NA67	NA67	NA27	NA67
-----% Sev. ----- Infection Type -----									
AC Assiniboia	Tr S	20 S	MS	80 S	4	4	4	2	4
Buff	60 S	60 S	S	60 S			4	4	4
Ebeltoft	5S	TS	MR-MS	20 S			4	2	4
HiFi	5 MR-S	5 MR-MS	MR-MS	20 S	?	;	3	1	2
Hytest	60 S	20 S	S	60 S	3		4	4	4
Jerry	5S	10 S	S		3		4	2	4
AC Kaufman				60 S					4
Killdeer	5 MR	10 S	MS		4		4	1	4
Ac Medallion	10 S	5 MR-S	S	80 S	4	4	4	1	4
Morton	5 S	5 S	MS	80 S	4	4	4	2	4
Otana	60 S	60 S	S	80 S	4		4	4	4
Paul	Tr R-MR	0R	R-MR	Tr R				1	;
Youngs	5 MR-MS	10 R-MS	MR-MS	60 S	4		4	1	4
Beach	60 S	20 S	MR-MS	60 S	3		4	2	4
Stark	Tr R	Tr R	R	Tr R-MR	?	;	;	;	;

19d. Exhibit D. Additional Description of the Variety

Table 7

2000-2003 Oat crown rust evaluation

	Fargo Field Results				Greenhouse Seedling Evaluations						
Genotype	2000	2002	2003		2000 Composite	2001			2002		2003 Comp
	Yield Plot	Yield Plot	YieldPlot	Hill Plot		Comp.	Hot CR	Comp.	Comp	Comp+	
----- % Sev. -----					----- Infection Type -----						
AC Assiniboia	OR		OR	10 MS	;	;	;	;	/1-4	;	;
Buff			10 MR-MS	40 MR-MS					4	3	3
Ebeltoft	20 MR-MS		10 MR-MS	40 MR-MS	/3-3?	4	4	4	4	3	4
HiFi	OR		OR	OR	/2-2	;	;	/1-4	/1-3	/1-3	;
Hyttest	40 MS		20 MR-MS	60 S	3	4	4	3	4	3	4
Jerry	60 MS	20 MR-MS	20 MR-MS	60 S	3	4	4	4	4	3	4
AC Kaufman			OR	OR							;
KILLDEER	40 MR-MS	40 MS	60 S	60 S	3	4	4	4	4	3	3
Loyal	OR		Tr R-MR	10 MR-MS	3	3	4	2	2	3	3
MEDALLION AC	OR		OR	OR	;	;	;	;	;	;	;
Monida	100 S	100 S	60 S	80 S	4	4	4	4	4	4	4
Morton	OR		OR	OR	;	;	;	;	;	;	/C
OTANA	100 S	80 S	80 S	100 S	3	4	4	4	4	4	4
PAUL	10 MR-MS		5 MS	20 MR	4	4	4	4	4	4	4
YOUNGS	40 MR-MS		20 S	40 MS	4	4	4	4	4	3	4
Beach	20 MR	20 MS	10 MR-MS	20 MR-MS	2	4	4	4	4	3	3
Stark	10 MR-MS		5 MR-MS	10 MR-MS	3	3	4	4	4	3	3

U.S. DEPARTMENT OF AGRICULTURE
AGRICULTURAL MARKETING SERVICE

Application is required in order to determine if a plant variety protection certificate is to be issued (7 U.S.C. 2421). The information is held confidential until the certificate is issued (7 U.S.C. 2426).

**EXHIBIT E
STATEMENT OF THE BASIS OF OWNERSHIP**

1. NAME OF APPLICANT(S) NDSU Research Foundation	2. TEMPORARY DESIGNATION OR EXPERIMENTAL NUMBER ND960736	3. VARIETY NAME 'Stark'
4. ADDRESS (Street and No., or R.F.D. No., City, State, and ZIP, and Country) C/O Executive Director PO Box 5002 Fargo, ND 58105-5002	5. TELEPHONE (Include area code) (701) 231-8931	6. FAX (Include area code) (701) 231-6661
7. PVPO NUMBER 200600022		

8. Does the applicant own all rights to the variety? Mark an "X" in the appropriate block. If no, please explain. ☒ YES ☐ NO

9. Is the applicant (individual or company) a U.S. national or a U.S. based company? If no, give name of country. ☒ YES ☐ NO

10. Is the applicant the original owner? ☐ YES ☒ NO If no, please answer one of the following:

a. If the original rights to variety were owned by individual(s), is (are) the original owner(s) a U.S. National(s)?

☒ YES ☐ NO If no, give name of country

b. If the original rights to variety were owned by a company(ies), is (are) the original owner(s) a U.S. based company?

☒ YES ☐ NO If no, give name of country

11. Additional explanation on ownership (Trace ownership from original breeder to current owner. Use the reverse for extra space if needed):

See additional Exhibit E Statement on the Basis of the applicant's ownership included in the application.

PLEASE NOTE:

Plant variety protection can only be afforded to the owners (not licensees) who meet the following criteria:

1. If the rights to the variety are owned by the original breeder, that person must be a U.S. national, national of a UPOV member country, or national of a country which affords similar protection to nationals of the U.S. for the same genus and species.
2. If the rights to the variety are owned by the company which employed the original breeder(s), the company must be U.S. based, owned by nationals of a UPOV member country, or owned by nationals of a country which affords similar protection to nationals of the U.S. for the same genus and species.
3. If the applicant is an owner who is not the original owner, both the original owner and the applicant must meet one of the above criteria.

The original breeder/owner may be the individual or company who directed the final breeding. See Section 41(a)(2) of the Plant Variety Protection Act for definitions.

According to the Paperwork Reduction Act of 1995, an agency may not conduct or sponsor, and a person is not required to respond to a collection of information unless it displays a valid OMB control number. The valid OMB control number for this information collection is 0581-0055. The time required to complete this information collection is estimated to average 0.1 hour per response, including the time for reviewing the instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information.

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19e. Exhibit E. Statement of the Basis of the Owner's Ownership

Dr. Michael S. McMullen, an employee of the North Dakota Agricultural Experiment Station and North Dakota State University, is a plant breeder who developed 'Stark' spring oat for which Plant Variety Protection is hereby sought. The employee by agreement and because of the condition of the use of facilities and funds of the North Dakota Agricultural Experiment Station and North Dakota State University has assigned all ownership rights to Stark oat to the North Dakota Agricultural Experiment Station and the North Dakota State University.

North Dakota State University on behalf of the North Dakota Agricultural Experiment Station has assigned all ownership to the NDSU Research Foundation. NDSU/RF is a nonprofit corporation set up to own and manage the intellectual property of North Dakota State University.